

D0134 NP

Figure 1A

1	CCAATAACAACCGGGGCGTTCACCTTCGGATTAGCAAGGTTTTGAAAACTATTTAGGT	60
61	GCCCATAAAAGGTCCCCTTCAGGTACCGGTCCGAAATTCCGGGTCAACCCAGGGTCCGGA	120
121	TCAATTAAGGGTCTGGGGGTGGCCACCTGGCCACTTTGGAAAATTGCAAGCATTTCCAA	180
181	GCTTCATCCGGCTCCAGGGTTGGCCTCTCCAAAAGGCAGGCGGCTTTTAAACGGGTCCA	240
241	ACAGAAAGGACCTCCCTTGGTCTCCTCAATTCCTGGCTGGAGTTTCTCTTCTCGTGTGT	300
301	GGAAGGATTCCAAACCCACACACAGGACCCGCATCCTGGGTGATGAAGTCAGACACGC	360
361	AGCAGCTGGGTGAGTGCTACGCTCAAGATAAGCATCTGTGCCATTGTGGGGACTCCCTGG	420
421	GCTGCTCTGCACCCGGACACTTGCTCTGTCCCGCCATGTACAACGGGTCGTGCTGCCGC	480
1	M Y N G S C R	8
481	ATCGAGGGGGACACCATCTCCCAGGTGATGCCGCCGCTGCTCATTGTGGCCTTTGTGCTG	540
9	I E G D T I S Q <u>V M P P L L I V A F V L</u>	28
541	GGCGCACTAGGCAATGGGGTCGCCCTGTGTGGTTTCTGCTTCCACATGAAGACCTGGAAG	600
29	<u>G A L G N G V A L C G F</u> C F H M K T W K	48
601	CCCAGCACTGTTTACCTTTTCAATTTGGCCGTGGCTGATTTCTCCTTATGATCTGCCTG	660
49	P S T <u>V Y L F N L A V A D F L L M I C L</u>	68
661	CCTTTTCGGACAGACTATTACCTCAGACGTAGACACTGGGCTTTTGGGGACATTCCCTGC	720
69	<u>P F</u> R T D Y Y L R R R H W A F G D I P Q	88
721	CGAGTGGGGCTCTTCACGTTGGCCATGAACAGGGCCGGGAGCATCGTGTTCCTTACGGTG	780
89	R <u>V G L F T L A M N R A G S I V F L T V</u>	108
781	GTGGCTGCGGACAGGTATTTCAAAGTGGTCCACCCCCACCACGCGGTGAACACTATCTCC	840
109	<u>V A A</u> D R Y F K V V H P H H A V N T I S	128
841	ACCCGGGTGGCGGCTGGCATCGTCTGCACCCTGTGGGCCCTGGTCATCCTGGGAACAGTG	900
129	T R V <u>A A G I V C T L W A L V I L G T V</u>	148
901	TATCTTTTGCTGGAGAACCATCTCTGCGTGCAAGAGACGGCCGTCTCCTGTGAGAGCTTC	960
149	<u>Y L L L</u> E N H L C V Q E T A V S Q E S F	168

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Figure 1B

961	ATCATGGAGTCGGCCAATGGCTGGCATGACATCATGTTCCAGCTGGAGTTCTTTATGCCC	1020
169	I M E S A N G W H D <u>I M F Q L E F F M P</u>	188
1021	CTCGGCATCATCTTATTTTGCTCCTTCAAGATTGTTTGGAGCCTGAGGCGGAGGCAGCAG	1080
189	<u>L G I I L F</u> <u>S F K I V W S L</u> R R R Q Q	208
1081	CTGGCCAGACAGGCTCGGATGAAGAAGGCGACCCGGTTCATCATGGTGGTGGCAATTGTG	1140
209	L A R Q A R M K K A T R <u>F I M V V A I V</u>	228
1141	TTCATCACATGCTACCTGCCCAGCGTGTCTGCTAGACTCTATTTCTCTGGACGGTGCCC	1200
229	<u>F I T C Y L P S V S A R L Y</u> F L W T V P	248
1201	TCGAGTGCCTGCGATCCCTCTGTCCATGGGGCCCTGCACATAACCCTCAGCTTCACCTAC	1260
249	S S A <u>D P S V H</u> <u>G A L H I T L S F T Y</u>	268
1261	ATGAACAGCATGCTGGATCCCCCTGGTGTATTATTTTCAAGCCCCTCCTTTCCCAAATTC	1320
269	<u>M N S M L D P L V Y Y F</u> S S P S F P K F	288
1321	TACAACAAGCTCAAAATCTGCAGTCTGAAACCAAGCAGCCAGGACACTCAAAAACACAA	1380
289	Y N K L K I C S L K P K Q P G H S K T Q	308
1381	AGGCCGGAAGAGATGCCAATTTCGAACCTCGGTGCGAGGAGTTGCATCAGTGTGGCAAAA	1440
309	R P E E M P I S N L G R R S C I S V A K	328
1441	GTTTCCAAAGCCAGTCTGATGGGCAATGGGATCCCCACTTGTTGAGTGGCACTGAACAAG	1500
329	V S K A S L M G N G I P T C	342
1501	CAGACCAACAACACTGAGGAAGATAGAGTGGTGACTTAGAATTAACTCGTGCTAAGGGGT	1560
1561	CGGGGGCTTTGAAAATGCCACCCCCCTTCTTATTGCAAGACGGCTTCTCGCACATGAAC	1620
1621	TGCATCCTTCTCATTCTGTTCGGAAATGAAATTACACAACCTATACCTTTTGGGGAGGTTC	1680
1681	CAGTTGATTGAAGTGAGTTGGCTGCATTTTCTTATCTGATCACAATGGCAGGGGACAGAA	1740
1741	TGTGCATGGAGTGGAGCATGTGTGTGTTGGGAGGGGGCTAGGAAGTGCACAGCCCTTGT	1800
1801	GTAATTTTCGTTGTTTGTGTTTGTGTTTGGAGACAGAGTCTCACTCTGTGTCCCAGGCTGGA	1860
1861	GTGCAGTGGCACAGTCTCGGCTCACTGCAACCTCTGCCTCCCGGGTTCAAGCAATTCTCC	1920
1921	TGCCTCAGCCTCCCGAGTAGCTGGGATTAGAGGCGCCAGCCAACACACCCGGCTAATTTT	1980

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Figure 1C

1981	TGTATTTT [.] TAGTAGAGACAGGGTTT [.] TGCCATGTTGGCCAGGCTGGTCT [.] CGAGCTCCTGAC [.]	2040
2041	CTCAGGTGATCCGCCTGCCTTGGCCTCCCAAGTGGTGGGATCACAGGCGTGAGCCACCG [.]	2100
2101	TGCCCCGGCCTCCCCTGTGT [.] CATTTTAAATGGCTAAGTAAATGGGTATATGTGTTGAATG [.]	2160
2161	GGGCATGTTCACTCTCTTAGGGGCTATGGGGCAGTTAGCAGCATTTCCCTATCCTCTGACC [.]	2220
2221	TTAAATCATTCCTTATCTCAGAAAACAGAAACCGGGCTCAGTCAATCAATGCTTTATTTT [.]	2280
2281	AGGCCGAATGAGGCTCTTTAGATTGGGATCTATTGATCTATCAATTTTCATCTTTACATT [.]	2340
2341	TCTTTGTACATCTGTACATTTTGTCCAAATGTACATCTGTACGTCTGTCATCATTGTGAC [.]	2400
2401	TTCTTGGTAGCCCAAGAAGAACAACAACAAAACAATCTGCTCTGACCTTCTTCAAATCTT [.]	2460
2461	TGTATTTCAAAGAAGGTGCTGAGGGATCTGTTTCCTTGCCCTGGCTTCTCCAGTGGGATG [.]	2520
2521	TGCTGAGTCCAATACAATTGCTTTTATAATTGCTTTTGA [.] AAAAAAAAAAAAAAAAAAAAAAG [.]	2580

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Figure 2A

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P2YR_CHICK MTEALISAALNGTQPELLAG..G..W.....AAGNATTKCSLTKTGFQFYFLPTVYIL
P2YR_MELGA MTEALISAALNGTQPELLAG..G..W.....AAGNASTKCSLTKTGFQFYFLPTVYIL
P2YR_BOVIN MTEVLWPAVFNGTDTAFLADPGSPWGNSTVTSTAASVSPFKCALTKTGFQFYFLPAVYIL
P2YR_HUMAN MTEVLWPAVFNGTDAFLADPGSSWGNSTVASTAAVSSSFKCALTKTGFQFYFLPAVYIL
P2YR_RAT MTEVPWSAVFNGTDAFLADGLSLWGNSTVASTAAVSSSFKCALTKTGFQFYFLPAVYIL
HGPRBMY27 ~~~~~MYNGSCCRIEGDTISQVMPPLIIV
HM74_HUMAN ~~~~~MNRHHLQDHFLEIDKKNCCVFRDDFIKAVLPPVLGL
GPRV_HUMAN ~~~~~MPFPNCSA.PSTVVATAVGVLLGL

P2YR_CHICK VFITGFLGNSVAIWMFVFHMPWSGISVYMFNLALADFLYVLTLPALIFYFNKTDWIFG
P2YR_MELGA VFITGFLGNSVAIWMFVFHMPWSGISVYMFNLALADFLYVLTLPALIFYFNKTDWIFG
P2YR_BOVIN VFITGFLGNSVAIWMFVFHMPWSGISVYMFNLALADFLYVLTLPALIFYFNKTDWIFG
P2YR_HUMAN VFITGFLGNSVAIWMFVFHMPWSGISVYMFNLALADFLYVLTLPALIFYFNKTDWIFG
P2YR_RAT VFITGFLGNSVAIWMFVFHMPWSGISVYMFNLALADFLYVLTLPALIFYFNKTDWIFG
HGPRBMY27 AFVLGALGNGVALCGFCFHMKTWKPSIVYLFNLAVADFLMLTCLPFRTDYYLRRRWAFG
HM74_HUMAN EFIFGLLGNGLALWIFCFHDKSKWSSRLFLNLAVADFLMLTCLPFRMDYYVRRSDWNEG
GPRV_HUMAN ECGGLLGNALVALWTFLEFRVRVMKPYAVYLLNLALADULLAACLPFLAAFYLSLQAWHLG

P2YR_CHICK DVMCKLQRFIFHVNLVGSILFLTCISVHRYTGVVHPLKSLGRLKKKNAYVYSSLVWALVV
P2YR_MELGA DVMCKLQRFIFHVNLVGSILFLTCISVHRYTGVVHPLKSLGRLKKKNAYVYSSLVWALVV
P2YR_BOVIN DAMCKLQRFIFHVNLVGSILFLTCISAHRYSGVVYPLKSLGRLKKKNAYVYSSLVWALVV
P2YR_HUMAN DAMCKLQRFIFHVNLVGSILFLTCISAHRYSGVVYPLKSLGRLKKKNAYVYSSLVWALVV
P2YR_RAT DVMCKLQRFIFHVNLVGSILFLTCISAHRYSGVVYPLKSLGRLKKKNAYVYSSLVWALVV
HGPRBMY27 DIPCRVGLFTLAMNRAGSIVFLTVMAADRYFKVVEHHAANTISTRTAAGIVCTEWALVV
HM74_HUMAN DIPCRVGLFTLAMNRAGSIVFLTVMAADRYFKVVEHHAANTISTRTAAGIVCTEWALVV
GPRV_HUMAN RVGCWALRFLDLSRSVGMATLAAMALDRYLRVHERLKNLSPQAALGVSGLVWALVV

P2YR_CHICK AVIAPILFYSGTGVRNKTTITCYDTTADEYLRSYFYISMCTTVFMFCIFFLVILGCYGLI
P2YR_MELGA AVIAPILFYSGTGVRNKTTITCYDTTADEYLRSYFYISMCTTVFMFCIFFLVILGCYGLI
P2YR_BOVIN VGISPILFYSGTGIRKNKTITCYDTTSDAYLRSYFYISMCTTVAMFCVPLVILGCYGLI
P2YR_HUMAN VAISPILFYSGTGVRNKTTITCYDTTSDAYLRSYFYISMCTTVAMFCVPLVILGCYGLI
P2YR_RAT VAISPILFYSGTGIRKNKTITCYDTTSDAYLRSYFYISMCTTVAMFCVPLVILGCYGLI
HGPRBMY27 LGTVYLLLENHL.CVQETAVSCESFI..MESANG..WHDIMFQLEFFMPLGITILFCSFKRI
HM74_HUMAN GLTVHLLKKKLL..QONGPANVCISFS..ICTHFR..WHEAMFLEFLPLGITILFCSFKRI
GPRV_HUMAN ALTCEGLLISEA.AQNSTR..CHSFYSRADGSFSIIMQEALSCLQFVLEFGLIVFCNAGI

P2YR_CHICK VKAL..IYKDLDNSPLRRKSIYLVIIIVLTVFAVSYPFHVMKTNLRLARLDFQTPQMCAF
P2YR_MELGA VKAL..IYKDLDNSPLRRKSIYLVIIIVLTVFAVSYPFHVMKTNLRLARLDFQTPQMCAF
P2YR_BOVIN VKAL..IYKDLDNSPLRRKSIYLVIIIVLTVFAVSYPFHVMKTNLRLARLDFQTPQMCAF
P2YR_HUMAN VKAL..IYKDLDNSPLRRKSIYLVIIIVLTVFAVSYPFHVMKTNLRLARLDFQTPQMCAF
P2YR_RAT VKAL..IYKDLDNSPLRRKSIYLVIIIVLTVFAVSYPFHVMKTNLRLARLDFQTPQMCAF
HGPRBMY27 VWSLRRR.QQLARQARMKKATRFIMVVAIVFITCYLP.SVSARLYFLWTVPSSA...CD.
HM74_HUMAN VWSLRQR..QMDRHAKIKRAITFIMVVAIVFITCYLP.SVVVRIRIFWLLHTSGTQNCVEV
GPRV_HUMAN IRLAQKRLREPEKQPKLQRAQALVTLVVVLFALCFLE.CFLARM.LMHIFQNLG..SCRA

P2YR_CHICK NDKVYATYQVTRGLASLNSCVDPILYFLAGDTFRRRLSRATRKSSRRSEPNVQSKSEEMT
P2YR_MELGA NDKVYATYQVTRGLASLNSCVDPILYFLAGDTFRRRLSRATRKSSRRSEPNVQSKSEEMT
P2YR_BOVIN NDRVYATYQVTRGLASLNSCVDPILYFLAGDTFRRRLSRATRKASRRSEANLQSKSEEMT
P2YR_HUMAN NDRVYATYQVTRGLASLNSCVDPILYFLAGDTFRRRLSRATRKASRRSEANLQSKSEEMT
P2YR_RAT NDRVYATYQVTRGLASLNSCVDPILYFLAGDTFRRRLSRATRKASRRSEANLQSKSEEMT
HGPRBMY27 .PSVHGALHTLSFTYMNMSLDPLVYVYFSSPSFPKFNKLIKCSLKP.K.QPGHSKTORPE
HM74_HUMAN YRSVDLAFITLSFTYMNMSLDPLVYVYFSSPSFPNFFSTLINECLQK.MTGEPDNNRST
GPRV_HUMAN LCAVAHTSDVTGSLTYLHSSVNVVYVYCFSSPTFRSSYRNVFH.IILRGKQAAEPPDFNPR

P2YR_CHICK LNILTEYKQNGDTS~~~~~
P2YR_MELGA LNILTEYKQNGDTS~~~~~
P2YR_BOVIN LNILSEFKQNGDTS~~~~~
P2YR_HUMAN LNILPEFKQNGDTS~~~~~
P2YR_RAT LNILSEFKQNGDTS~~~~~
HGPRBMY27 EMPISNLGRRSCISVAKVSKASLMGN.GIPTC~~~~~
HM74_HUMAN SVELTGDPNKT....RGAPEALMANSGEPSYLGPTSNNHKKGHCHQEPASLEKQL
GPRV_HUMAN DSYST~~~~~

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Figure 2B

P2YR_CHICK ~~~~~
P2YR_MELGA ~~~~~
P2YR_BOVIN ~~~~~
P2YR_HUMAN ~~~~~
P2YR_RAT ~~~~~
HGPRBMY27 ~~~~~
HM74_HUMAN GCCIE
GPRV_HUMAN ~~~~~

Figure 3

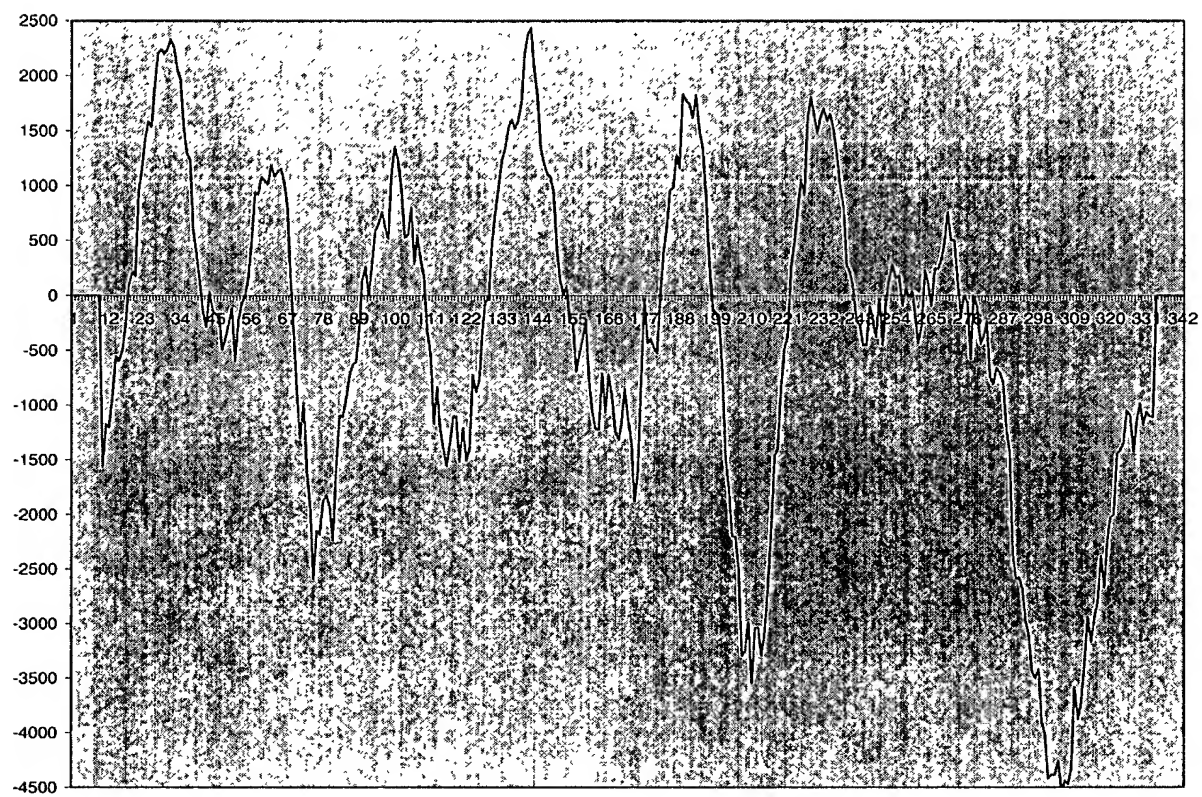
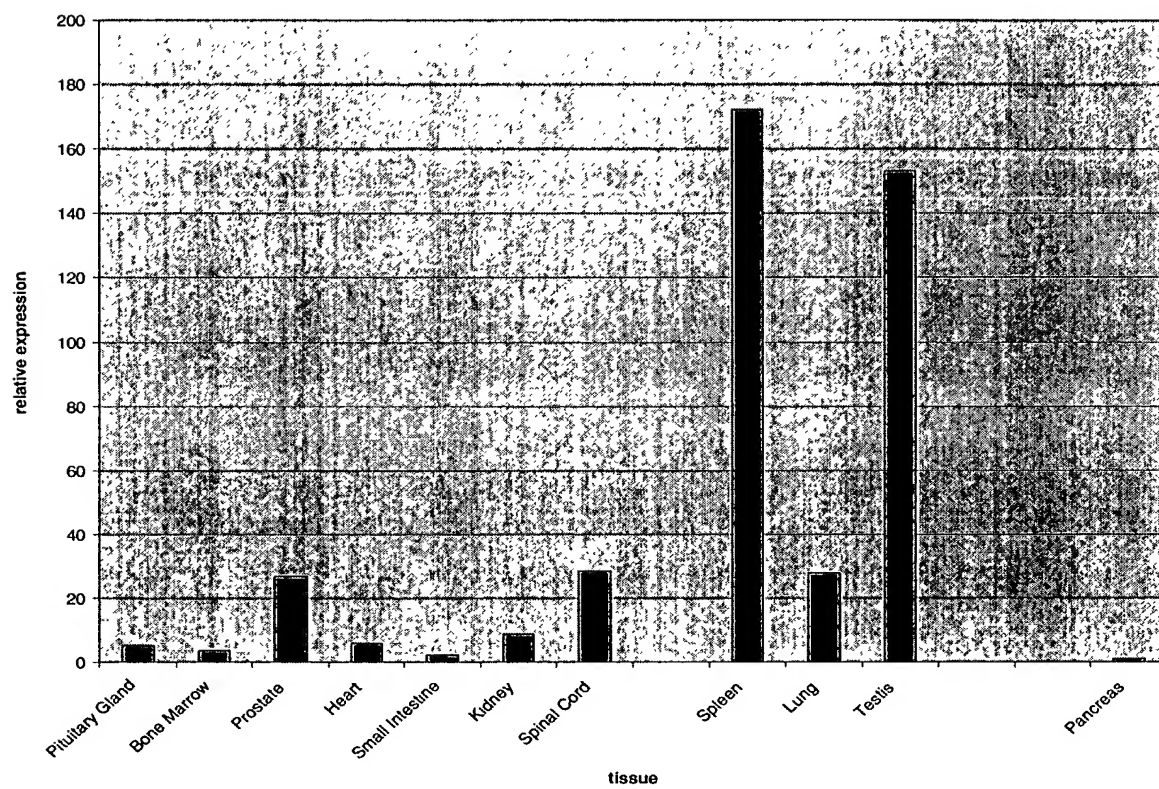


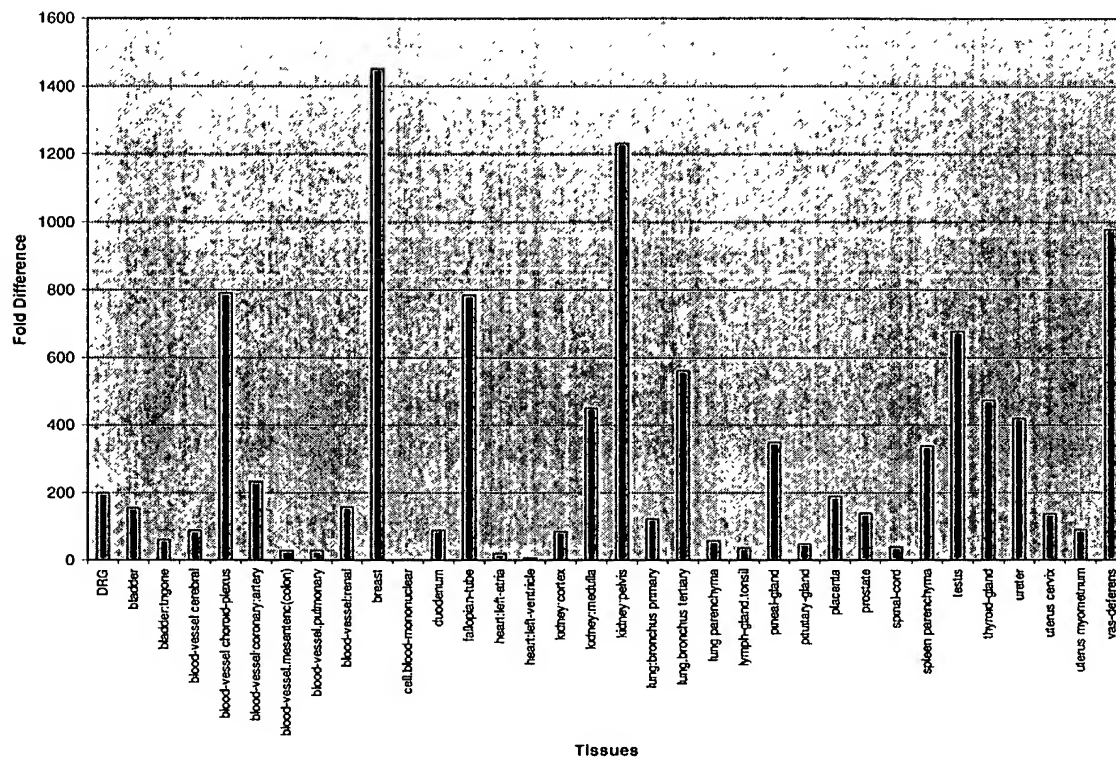
Figure 4

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Figure 5.

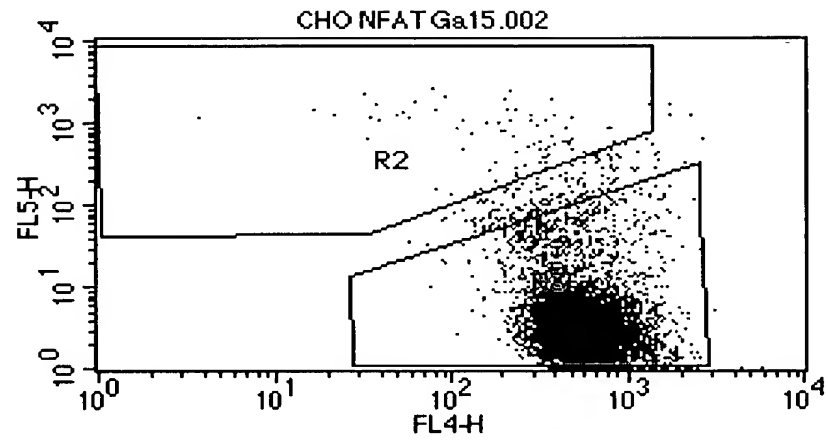
<u>Protein</u>	<u>SWISS- PROT ID</u>	<u>Identities</u>	<u>Similarities</u>
chicken P2Y purinoceptor 1 (ATP RECEPTOR) protein	P34996	28.9%	39.4%
turkey P2Y purinoceptor 1 (ATP RECEPTOR) protein	P49652	28.9%	39.4%
bovine P2Y purinoceptor 1 (ATP RECEPTOR) protein	P48042	28.7%	39.8%
human P2Y purinoceptor 1 (ATP RECEPTOR) protein	P47900	28.7%	39.5%
rat P2Y purinoceptor 1 (ATP RECEPTOR) protein	P49651	28.7%	39.8%
human G protein-coupled receptor, HM74 protein	P49019	53.6%	61.0%
human G protein-coupled receptor, GPR31 protein	O00270	33.0%	43.6%

Figure 6



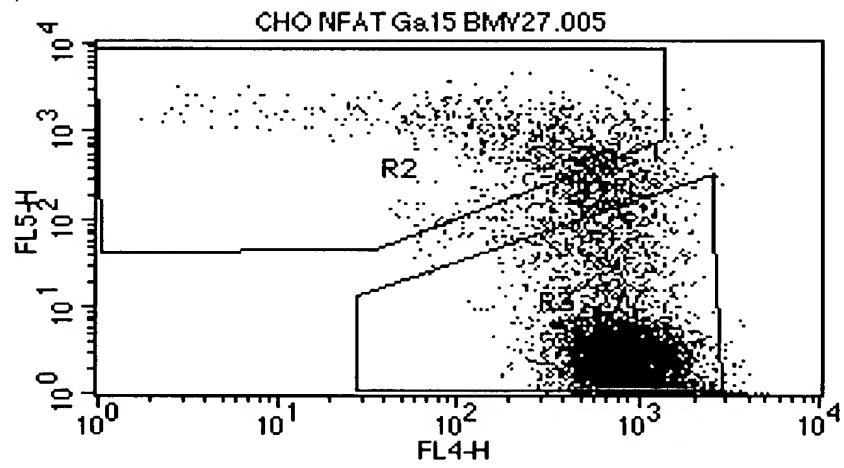
D0134 NP

Figure 7



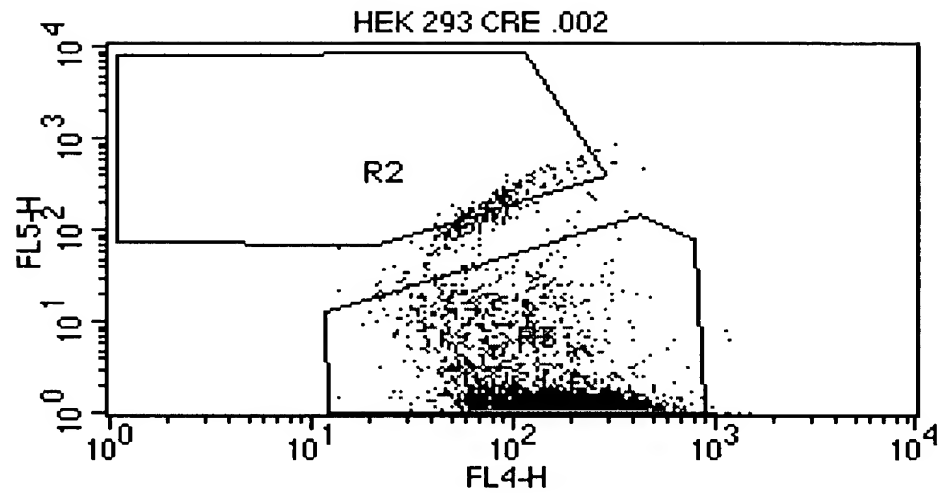
D0134 NP

Figure 8



D0134 NP

Figure 9



D0134 NP

Figure 10

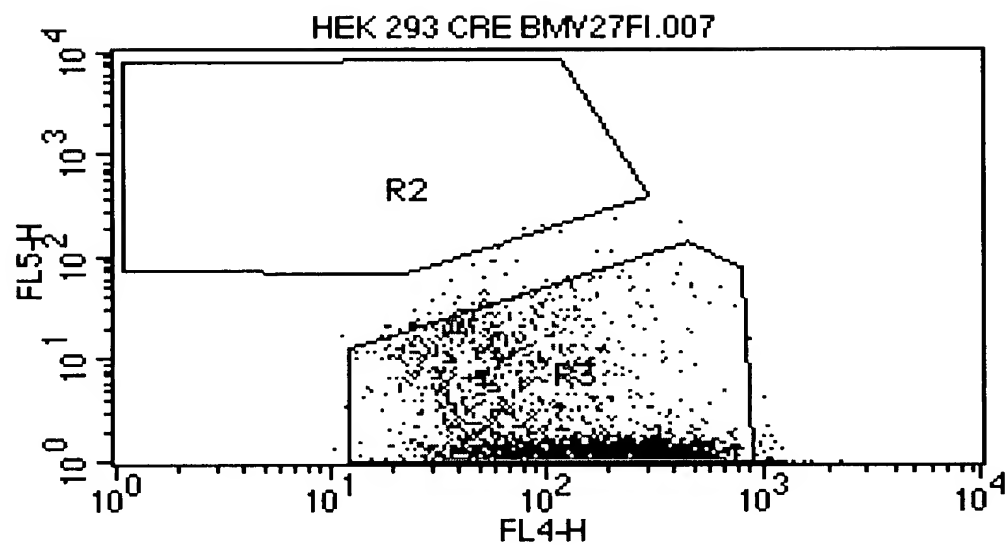
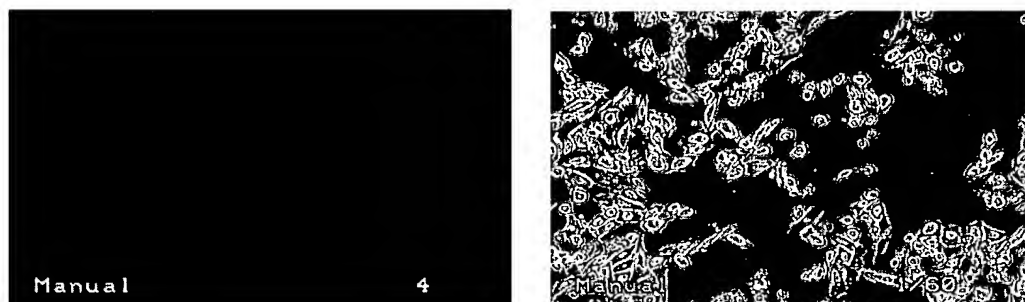


Figure 11

Cho NFAT Ga15 Control (Fluorescent vs. Bright Field)



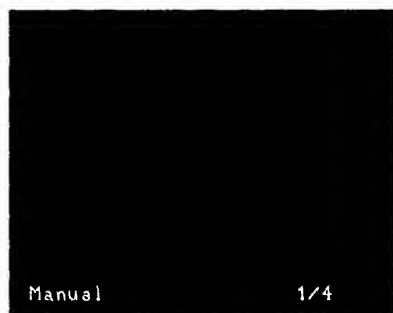
Cho NFAT Ga15 BMY27 (Fluorescent vs. Bright Field)



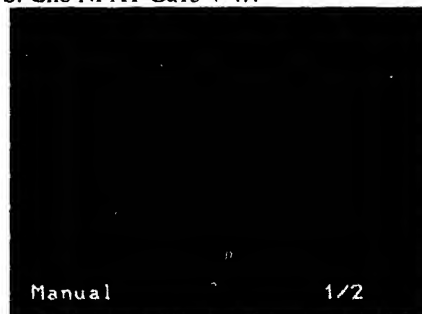
D0134 NP

Figure 12

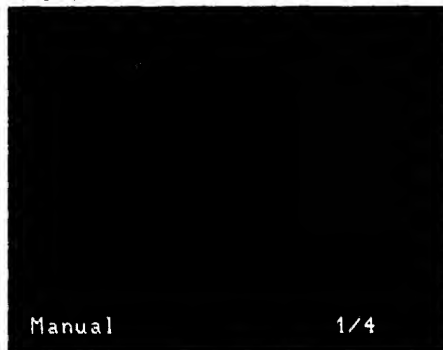
a. Cho NFAT Ga15



b. Cho NFAT Ga15 + T/P



c. Cho NFAT Ga15 oGPCR-Intermediate



d. Cho NFAT Ga15 oGPCR High

